REMARKS

Reconsideration is respectfully requested in light of the foregoing amendments and remarks, which follow.

Claims 51-64 are now before the Examiner for consideration. These claims are directed to a screening method and require the presence of the N-boxes. There are inherent in the system as claimed, N-boxes foreign to the cells present in the cell and also those normal to the cells.

Claims 26 through 45 and 51-64 have been withdrawn from consideration by the Examiner under 37 CFR 1.141(b) as directed to non-elected subject matter. Claims 1-25 and 46-50 are cancelled. No new matter is deemed to have been added by the newly submitted claims.

This response supplements that filed on November 13, 2002, the content of which is expressly incorporated herein by reference. It is noted that the Office considered that response in an Official Action dated March 4, 2003.

Objection to the Sequence Listing

The application was objected to for failing to comply with the requirements of 37 CFR 1.821 through 1.825. On June 4, 2002, the Applicant resubmitted a complete paper copy of the sequence listing and a copy in computer readable form in the Response filed November 13, 2002. It is noted that in the Office communication dated March 4, 2003 that the disc was damaged, a new one was requested. The instant application was recently transferred to this firm. A new disc will be submitted shortly.

Rejection under the second paragraph of 35 U.S.C. §112

Claims 46-50 are rejected under 35 U.S.C. 112, second paragraph as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claims 46-50 have been cancelled and new claims 51-64 have been added. These claims were prepared in accordance with the Examiner's suggestions.

Withdrawal of the rejection is respectfully requested.

Rejections under 35 U.S.C. 112, first paragraph

Claims 46-50 were rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor, at the time the application was filed, had possession of the claimed invention. Applicant respectfully traverse.

Claims 51-64 are directed to screening methods for compounds that modify the copy number of a cis-acting element, specifically, N-boxes, that can bind the limiting cellular transcription complex GABP•p300/cbp.

The claimed method relates to the introduction of a compound into a system containing active N-boxes and measuring the compound's effect on the N-box interaction with the preexisting components in the system. The disclosed invention does not lie in the development of new assay components or methodology, but rather in the new use of existing assay components to measure the effect of a suspect compound on a the newly discovered relationship between decreased availability of a limiting cellular transcription complex and disease. The content of the specification describes the discovery and recognition of the relation ship and describes the operation of the assay. Consider the following summary of the discovery.

Many cellular genes bind GABP

Many cellular genes are known which bind GABP. GABP binds promoters and enhancers of many cellular genes including, interleukin 16 (IL-16), interleukin 2 (IL-2), interleukin 2 receptor β-chain (IL-2Rβ), IL-2 receptor γ-chain (IL-2 γc), human secretory interleukin-1 receptor antagonist (secretory IL-1ra), retinoblastoma (Rb), human thrombopoietin (TPO), aldose reductase, neutrophil elastase (NE), folate binding protein (FBP), cytochrome c oxidase subunit Vb (COXVb), cytochrome c oxidase subunit IV, mitochondrial transcription factor A (mtTFA), β subunit of the FoF1 ATP synthase (ATPsynβ), prolactin (prl) and the oxytocin receptor (OTR). For some of these genes, for instance, CD18, COXVb, COXIV, GABP binds to the promoter while for others, for example IL-2 and ATPsynβ GABP binds an

enhancer. Specifically, GABP binds retinoblastoma susceptible gene (Rb) (see section entitled "Retinoblastoma susceptible gene (Rb)," p 31 line 26 to p 32 line 12 in application), breast cancer type 1 gene (BRCA1) (see section entitled "Breast cancer type 1 gene (BRCA1)," p 32 line 13-26 in application), Fas gene (Fas, APO-1, CD95) (see section entitled "Fas gene (Fas, APO-1, CD95)," p 33 line 1-17 in application), tissue factor (TF) gene (see section entitled "Tissue factor (TF)," p 33 line 18 to p 39 line 7 in application), P-selectin gene (see section entitled "P-selectin gene," p 41 line 28 to p 42 line 26 in application), β_2 leukocyte integrin (CD18) (see sections entitled "β_2 integrin gene," p 43 line 1-8 in application), α_4 integrin (CD49d) (see section entitled "α_4 integrin gene," p 43 line 9-21 in application), and hormone sensitive lipase (HSL) gene (see section entitled "Hormone sensitive lipase (HSL) gene," p 43 line 22 to p 45 line 17 in application).

Viral DNA also binds GABP

The cis-acting element that binds GABP is known as the N-box. The N-box is the core binding sequence of many viral enhancers including the polyomavirus enhancer area 3 (PEA3), adenovirus E1A enhancer, Rous Sarcoma Virus (RSV) enhancer, Herpes Simplex Virus 1 (HSV-1) (in the promoter of the immediate early gene ICP4), Cytomegalovirus (CMV) (IE-1 enhancer/promoter region), Moloney Murine Leukemia Virus (Mo-MuLV) enhancer, Human Immunodeficiency Virus (HIV) (the two NF-κB binding motifs in the HIV LTR), Epstein-Barr virus (EBV) (20 copies of the N-box in the +7421/+8042 oriP/enhancer) and Human T-cell lymphotropic virus (HTLV) (8 N-boxes in the enhancer and one N-box in the LTR). Note that some viral enhancers, for example SV40, lack a precise N-box, but still bind the GABP transcription factor.

Ample evidence exists supporting binding of GABP to the N-boxes in these viral enhancers, for instance, binding of GABP to the HIV LTR, binding of GABP to the promoter of ICP4 of HSV-1, binding of GABP to the adenovirus E1A enhancer element I, binding of GABP (called EF-1A in their paper) to the polyomavirus enhancer and binding of GABP to Mo-MuLV. Studies also demonstrated competition between the above viral enhancers and enhancers of other viruses, e.g. competition between the Moloney Sarcoma Virus (MSV) enhancer and SV40 enhancer and competition between the RSV enhancer and the BK virus enhancer. Note, for example, "Viral GABP enhancers" on p 11 line 15 to p 12 line 11 in the application.

The GABP•p300/cbp transcription complex is limiting

The coactivator p300 is a 2,414-amino acid protein initially identified as a binding target of the E1A oncoprotein. cbp is a 2,441-amino acid protein initially identified as a transcriptional activator bound to phosphorylated cAMP response element (CREB) binding protein (hence, cbp). p300 and cbp share 91% sequence identity and are functionally equivalent. Both p300 and cbp are members of a family of proteins collectively referred to as p300/cbp.

Although p300/cbp are widely expressed, their cellular availability is limiting. Several studies demonstrated inhibited activation of certain transcription factors resulting from competitive binding of p300/cbp to other cellular or viral proteins. For example, competitive binding of p300, or cbp, to the glucocorticoid receptor (GR), or the retinoic acid receptor (RAR), inhibited activation of a promoter dependent on the AP-1 transcription factor. Competitive binding of cbp to STAT1α inhibited activation of a promoter dependent on both the AP-1 and ets transcription factors. Competitive binding of p300 to STAT2 inhibited activation of a promoter dependent on the NF-κB RelA transcription factor. Other studies also demonstrated that p300/cbp is limiting.

GABP binds p300/cbp. Since p300/cbp is limiting, the transcription complex GABP•p300/cbp is also limiting. Note also, for example in the instant application, the sections entitled "GABP binds p300," "Cellular availability of p300 is limited," and "Microcompetition for GABP•p300," on p 11 line 1-4, p 11 line 5-14, and p 12 line 12-21, respectively.

Infection with a GABP virus disrupts transcription of cellular GABP regulated genes, which, in turn, leads to disease

Consider an infection with a virus that binds GABP•p300/cbp. Does such infection disrupt cellular gene transcription? Take as example the BRCA1 gene. BRCA1 binds GABP•p300/cbp. Binding of GABP•p300/cbp transactivates the gene and increases concentration of the BRCA1 protein. GABP•p300/cbp is limiting. By binding GABP•p300/cbp, the virus decreases availability of the complex to the BRCA1 promoter. Decreased availability decreases binding of GABP•p300/cbp to the promoter, which, in turn, decreases BRCA1 transcription. Decreased BRCA1 expression increases the probability of

developing disease, specifically, breast and ovarian cancer (note also, for example, the instant application the sections entitled "Breast cancer type 1 gene" on p 32 and "BRCA1" on p 52-53).

In addition to BRCA1, the specification includes other cellular genes, Rb, Fas, HSL, TF, P-selectin, and α4 integrin, etc., susceptible to such disruption. As with BRCA1, for each of the other genes, the specification describes the effect of an infection with a GABP virus on gene transcription, cell function, and the clinical conditions of the organism infected by the virus.

The Applicant's disclosure as originally filed reasonably conveys to one skilled in the art that the Applicant had possession of the invention claimed in new claims 51-64. Therefore, reconsideration and withdrawal of the rejection is respectfully requested.

Claims 46-50 were rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention. Applicant respectfully disagrees.

Screening method

New claims 51-64 have been submitted. The new claims center on copy number assays and require the presence of N-boxes. The requisite steps and conditions of the screening method are set forth. Operation of the screening method relative to a compound of interest is described in the specification. Copy number assays are well known in the art. Moreover, the specification provides multiple examples of these assays such that one of ordinary skill in art would be able to practice the claimed invention.

It is not clear how undue experimentation is involved under the circumstances since the relationship under consideration is described and the components of system, where the relationship is thought to exist, are named. Measured changes are ascribed to the compounds activity.

The specification teaches at least three assays, routinely used to measure copy number of viral DNA. Consider the section in the application entitled: "Viral N-box agents." The section presents the LeBlanc 1999 study (present application, page 137, lines 15-23). The study

quantified the copy number of latent herpes simplex virus 1 (HSV-1) DNA using real-time PCR. Specifically, the study used the TaqMan system, ABI Prism 7700 Sequence Detector, which is the standard equipment in the industry (see Materials and Methods section in the paper). The Material and Method section includes the specific protocol used by LeBlanc, et al., to perform the real-time PCR assay.

The "Viral N-box agents" section also presents the Steffens 1998 study (present application, page 136, line 9 to page 137, line 2). The study quantified the number of cytomegalovirus DNA copies by performing serial dilutions of DNA, followed by PCR amplification (see Material and Method section in the paper). The Material and Method section includes the specific protocol used by Steffens, et al., to perform the quantitative viral DNA assay. Note that the study specifically measures the effect of a test therapy on the number of cytomegalovirus DNA copies, that is, measures the number of viral DNA copies before, and after contacting the subjects with the test therapy.

The "Viral N-box agents" section also presents the Bruisten 1998 study (present application, page 139, line 25 to page 140, line 19). This study measured the number of proviral HIV-1 DNA copies in PBMC using a competitive quantitative DNA PCR (see p 1054 in the paper). The paper includes the specific protocol used by Bruisten, et al., to perform the competitive quantitative DNA PCR assay.

These papers show working examples of exemplary assays. Moreover, the Material and Method sections of these papers, which include protocols for the specific assays, provide sufficient guidance to one reasonably skilled in the art on how successfully assay the copy number of viral DNA. Note that, since these three assays are well known in the art, there was not need to include the protocols in the specification.

Many assays are well known in the art for measuring the copy number of viral DNA (see three examples above). One such assay, real-time PCR, was routinely performed in laboratories around the world at the time the application was filed. The assay was first performed by Higuchi, *et al.*, as reported in their 1992 and 1993 papers (Higuchi R, Dollinger G, Walsh PS, Griffith R., *Simultaneous amplification and detection of specific DNA sequences*, Biotechnology (NY), 1992 Apr; 10(4):413-7 and Higuchi R, Fockler C, Dollinger G, Watson R. *Kinetic PCR*

analysis: real-time monitoring of DNA amplification reactions, Biotechnology (NY). 1993 Sep; 11(9): 1026-30), and quickly became the gold standard for the measurement of viral DNA copy number. A copy of these two articles is attached. At the time the application was filed, real-time PCR, was considered routine, easy to use, sensitive, accurate, reliable, and was extensively performed in academic settings and in pharmaceuticals companies. Guidance on how to perform real-time PCR was readily available. Many working examples existed showing successful measurement of viral DNA copy number using real-time PCR.

Since copy number assays are well known in the art and the components of the system to be measured is defined. It is not clear how undue experimentation is involved in the practice of the claimed method.

Withdrawal of the rejection as to enablement of the screening method is respectfully requested.

Specific Diseases

The Examiner states: "The specification fails to teach any evidence that the specific diseases recited in claims 47-50 or any other disease associated with decreased cellular availability of GABP" (office action p 6 line 4).

The specification presents microcompetition, a newly discovered relation between foreign polynucleotides, specifically, viral N-boxes, and cellular gene transcription. As a test of the new relation, the specification includes logical conclusions that describe the predicted effects of such relation on specific cellular gene transcription, cell function, and clinical condition of the organism harboring the foreign polynucleotides. The logical predictions are compared to observations reported in the literature. In all cases, the observations are consistent with the predicted effects, and therefore, validate the newly discovered relationship.

Note that although all the hundreds of cited studies used vastly different materials and methods, such as different foreign polynucleotides, different genes, different plasmids, different transfection methods, different cell types, and different organisms. The observations in all of the studies are still consistent with the predicted effects of microcompetition.

The benefit of large sample sizes, randomization, independent verification by different laboratories, etc, is to even out specific peculiarities inherent in any single measurement (see office action p 6 line 6 to p 7 line 6). Same results under dissimilar conditions are considered reliable. Since the predicted effects of microcompetition on specific gene transcription, cell function, and clinical conditions were confirmed under a variety of conditions using vastly dissimilar material and methods, the effects are not an artifact of any specific study, and therefore, highly reliable.

Bioinformatics is the study of relationships between single pieces of data. The present invention is based on a recognized level of scientific advancement. The present invention proposes a new relationship between microcompetition and disease and then tests the relationship against available evidence. Specifically, the following pieces of evidence support the new relationship:

- Microcompetition and its molecular effects (present application, pages 1-45)
- Clinical effects of Microcompetition- Cancer (present application, pages 45-57)
- Clinical effects of Microcompetition- Atherosclerosis (present application, pages 58-91)
- Clinical effects of Microcompetition- Osteoarthritis (present application, page 91-96)
- Clinical effects of Microcompetition- Obesity (present application, page 96-142).

Note that the individual pieces of evidence in the specification are published in the prior art, but the relationship was not. The conceptual difficulty in evaluating the present invention is the timing of the hypothesis and the experiments. Traditional schools teach aspiring empiricists to first state their hypothesis and then conduct experiments to test it. However, today so many experiments have been run and so much information is available that it is possible to suggest new hypotheses and then evaluate them against a multitude of already published observations. The present invention teaches the relationship between microcompetition and disease, and supports those teachings with numerous examples. The fact that the new relationship is supported by so many individual observations increases the reliability of the discovery.

The specification enables one of ordinary skill in the art to understand the relationship between microcompetition and disease and further to construct the claimed assays from known

components. In light of the above, it is respectfully submitted that the invention would be recognized as credible by one of skill in the art. Withdrawal of the rejection is respectfully requested.

Conclusion

Having addressed all the objections and rejections, the application is believed to be in condition for allowance and a notice to that effect is respectfully requested.

In the unlikely event that the transmittal letter is separated from this document and the Patent Office determines that an extension and/or other relief is required, applicants petition for any required relief including extensions of time and authorize the Director to charge the cost of such petitions and/or other fees due in connection with the filing of this document to Deposit Account No.22-0261 referencing docket no. 40630-188500. However, the Commissioner is not authorized to charge the cost of the issue fee to the Deposit Account.

Respectfully submitted,

Dated: July 18, 2003

By: 

Thomas G. Wiseman
Registration No. 35,046
VENABLE
P.O. BOX 34385
Washington, DC 20043-9998
Telephone: (202) 513-4614
Facsimile: (202) 962-8300